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Caracterização de nanopartículas de prata e avaliação do seu efeito antimicrobiano sobre *Salmonella*

Characterization of silver nanoparticles and evaluation of their antimicrobial effect on *Salmonella*

Caracterización de nanopartículas de plata y evaluación de su efecto antimicrobiano sobre *Salmonella*

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Resumo

Atualmente o sucesso da nanotecnologia atinge várias áreas da ciência, medicina, da tecnologia e, principalmente, do setor alimentício. As nanopartículas de prata (Ag-NPs) ganham destaque com seu efeito antimicrobiano que além de altamente tóxicas à celulas bacterianas, são facilmente sintetizadas e caracterizadas. O gênero *Salmonella* é o segundo maior causador de doenças e já existem relatos de sorotipos altamente resistentes. Assim, o uso de Ag-NPs é uma alternativa para controle bacteriano em alimentos. O objetivo deste

estudo foi sintetizar, caracterizar e verificar a atividade antimicrobiana de Ag-NPs sobre sorotipos de Salmonella. O tamanho das Ag-NPs foi estimado e foi possível detectar duas populações de 4,7 ± 0,09 e 35,7 ± 2,12. O potencial zeta foi de -33,7 ± 11,8 mV, indicando boa estabilidade da dispersão. A atividade antimicrobiana das Ag-NP foi determinada a partir da concentração inibitória mínima (CIM). A CIM mais baixa encontrada foi de 4,7 µg • mL-1 para *Salmonella* Enteritidis e a maior foi de 27,7 µg • mL-1 para o isolado de *Salmonella* Infantis 1. O uso de Ag-NPs é promissor em relação à atividade antimicrobiana, no entanto, melhorias nos métodos de síntese devem ser exploradas para viabilizar o uso comercial.

Palavras-chave: Nanopartículas de prata; Antimicrobianos; Indústria de alimentos.

Abstract

Currently, the success of nanotechnology affects several areas of science, medicine, technology and, especially, the food industry. Silver nanoparticles (Ag-NPs) stand out, with their antimicrobial effect. *Salmonella* is a cause of foodborne diseases and there are reports of resistant serotypes. The use of Ag-NPs is an alternative for bacterial control in foods. The aim of this study was synthesize, characterize and verify the antimicrobial activity of Ag-NPs on serotypes of *Salmonella*. The size of Ag-NPs was estimated and it was possible to detect two populations of 4.7 ± 0.09 and 35.7 ± 2.12. The zeta potential was -33.7 ± 11.8 mV indicating good dispersion stability.Ag-NP antimicrobial activity was determined from minimum inhibitory concentration (MIC).The lowest MIC found was 4.7 µg·mL^{-1} for *Salmonella* Enteritidis and the highest was 27.7 µg·mL^{-1} for *Salmonella* Infantis 1 isolate.The use of Ag-NPs is promising with respect to antimicrobial activity, however, improvements in synthesis methods should be explored in order to make commercial use viable.

Keywords: Silver nanoparticles; Antimicrobials; Food industry.

Resumen

Actualmente, el éxito de la nanotecnología afecta varias áreas de la ciencia, la medicina, la tecnología y, especialmente, la industria alimentaria. Se destacan las nanopartículas de plata (Ag-NP), con su efecto antimicrobiano. La salmonella es una causa de enfermedades transmitidas por los alimentos y hay informes de serotipos resistentes. El uso de Ag-NP es una alternativa para el control bacteriano en los alimentos. El objetivo de este estudio fue sintetizar, caracterizar y verificar la actividad antimicrobiana de Ag-NP en los serotipos de Salmonella. Se calculó el tamaño de Ag-NP y fue posible detectar dos poblaciones de 4.7 ± 0.09 y 35.7 ± 2.12. El potencial zeta fue de -33.7 ± 11.8 mV, lo que indica una buena

estabilidad de la dispersión. La actividad antimicrobiana Ag-NP se determinó a partir de la concentración inhibitoria mínima (MIC). La MIC más baja encontrada fue de 4.7 µg • mL-1 para Salmonella Enteritidis y la más alta fue de 27.7 µg • mL-1 para el aislado de Salmonella Infantis 1. El uso de Ag-NPs es prometedor con respecto a la actividad antimicrobiana, sin embargo, se deben explorar mejoras en los métodos de síntesis para hacer viable el uso comercial.

Palabras clave: Nanopartículas de plata; Antimicrobianos; Industria de alimentos.

1. Introduction

The genus *Salmonella*, belonging to the family *Enterobacteriaceae*, is composed of two species: *Salmonella enterica* and *Salmonella bongori*. The species *Salmonella enterica* is divided into six subspecies and the most commonly related to infections in animals and humans is *Salmonella enterica* subspecies *enterica* (Pui et al., 2011). This subspecies has more than 1500 serotypes, most of which are related to food-borne diseases with approximately 1.2 million cases of illness and 450 deaths per year (Steenackers et al., 2012; CDC, 2018).

Bacteria of the species *Salmonella enterica* are considered invasive intracellular pathogens, with the ability to colonize a range of animal or human hosts (Wagner & Hensel, 2011). The pathogenicity of *Salmonella enterica* results from its virulence, which is mediated by its high level of invasiveness, great capacity for intracellular proliferation and the production of several virulence factors such as endotoxins, enterotoxins and cytotoxins which can damage and kill the host cell (Peng, 2016; Madigan et al., 2015).

Use of antibiotics as bactericidal agents has been shown to have negative effects, such as high bacterial capacity to develop resistance and transfer resistance genes to other bacteria through plasmids, which is considered a major public health risk (Omara, Zawrah & Samy, 2017). Therefore, new treatments need to be studied as an alternative to antibiotic use.

In recent years, research involving nanotechnology has advanced in several areas of knowledge. In particular, Ag-NPs have gained importance as they present rapid methods of synthesis and characterization. Furthermore, they show antimicrobial activity against several microorganisms (Devi et al., 2017; Raman et al., 2017; Omara et al., 2017). The use of silver-containing products is considered an alternative for controlling the emergence of resistant bacteria (He & Hwang, 2016). In the last decade, several products containing silver and other nanoparticles have been approved by the FDA for use in food disinfection procedures and for

medical application (FDA, 2015). In fact, silver nanoparticles have shown great potential as an antimicrobial agent due to its bactericidal properties. The action mechanism of Ag-NPs is related to their characteristics of size, load and surface area (He & Hwang, 2016; Devi et al., 2017). Silver ions can form pores in the cell membrane, alter its permeability, catalyze the generation of reactive oxygen species (ROS) and lead the bacterial cell to die from oxidative stress. They may also interact with nitrogenous DNA bases preventing replication. Thus, the various action mechanisms involved may hinder the bacterial cell from activating resistance mechanisms (He & Hwang, 2016; Raman et al., 2017) or even deactivate it definitively. Thus, Ag-NPs can be considered as a potential alternative in the formulation of new antibacterial agents. Therefore, this study aimed to synthesize, characterize and evaluate the antimicrobial activity of Ag-NPs on different strains of *Salmonella* spp.

2. Methodology

The study was an experimental, quantitative research (Pereira, et al., 2018), which was carried out at the Laboratory of Microbiology of Food and Waterborne Pathogens (LAMPOAH), Department of Food Technology (DTA) belonging to the Universidade Federal de Viçosa (UFV), Minas Gerais.

2.1. Microorganisms

The microorganisms used in the evaluation of the antimicrobial activity of the Ag-NPs are described in Table 1.

Table 1: Bacteria used in the study.

Bacteria	Origin
Salmonella enterica subsp. *enterica* serotype Enteritidis (CT)	ATCC 13076*
Salmonella enterica subsp. *enterica* serotype Typhimurium	ATCC 14028*
Salmonella enterica subsp. *enterica* serotype Schwarzengrund	Isolated from Chicken
Salmonella enterica subsp. *enterica* serotype Heidelberg	Isolated from Chicken
Salmonella enterica subsp. *enterica* serotype Infantis	Isolated from Refrigerator
Salmonella enterica subsp. *enterica* serotype Infantis	Isolated from Refrigerator

*ATCC: *American Type Culture Collection* (EUA) given by Fundação Oswaldo Cruz (Fiocruz).

The Salmonella serotypes used in this study were isolated from different sources, the first two being from reference microorganisms (Fiocruz) and the others belonging to LAMPOAH.

2.2. Synthesis and preparation of Ag-NP solution

The Ag-NP solution was obtained by chemical reduction with sodium citrate according to the methodology described by Turkevich, Stevenson and Hillier (1951) with adaptations. In a beaker, 1000 mL of silver nitrate ($AgNO_3$) (SYNTH) 1 mmol·L^{-1} diluted in ultrapure water was heated. After two minutes of boiling, 10 mL of sodium citrate ($Na_3C_6H_5O_7$) (NEON) 36 mmol·L^{-1} was added and the solution was boiled for a further 4 min. Subsequently, the heat source was switched off and the end point was observed by a change in coloration to pale yellow.

2.3. Physical-chemical characterization of the silver nanoparticles

2.3.1. Determining the concentration of silver nanoparticles

The dispersed silver concentration was determined by an inductively coupled plasma optical emission spectrometer (ICP-OES) (Optima 8300 ICP-OES Spectrometer, PERKIN ELMER). In a specific tube, 1 mL of the Ag-NP solution was inserted and the intensity of the radiation emitted by the silver ions was measured. The data were compared with an analytical curve for silver (SIGMA-ALDRICH) at concentrations ranging from 0.1 to 5.0 µg·mL^{-1}. Ag-NP dispersion was appropriately diluted in deionized water for measurement in the apparatus.

2.3.2. Size and stability of silver nanoparticles (Dynamic Light Scattering (DLS) and Zeta Potential)

Zeta potential measurements were made using Zetasizer equipment (Zetasizer Nano-ZS, Malvern Instruments, Southborough, UK). In a specific cuvette 1 mL of the Ag-NP dispersion was inserted for reading the equipment. The dispersion was placed in electrophoresis and subjected to an electric field. The electrophoretic mobility was denominated by the speed at which the charged particles migrated to the opposite charge electrode with a velocity proportional to the potential (Malvern instruments, 2013). The zeta potential was calculated using Zetasizer Software data analysis software coupled to the equipment. The zeta potential values were determined in triplicate.

The hydrodynamic radius and polydispersity index of the Ag-NPs was also determined using Zetasizer equipment (Zetasizer Nano-ZS, Malvern Instruments, Southborough, UK).

The measurements were taken using an avalanche photodiode detector (Brookhaven BI-APD, USA) and a correlator (TURBOCORR, Brookhaven, USA). The light source (CVI Melles Griot, USA) was a HeNe laser of 35 mW of power and λ = 632.8 nm, linearly polarized. For intensity control, a system of crossed polarizers was used.

Particle size was defined as mean volumetric diameter (Σ Σ) and each result was automatically calculated by the software as the average of three measurements. After the size definition using DLS the solution dispersion index could be determined using Equation 1.

$$\mathbf{PDI} = \left(\frac{\boldsymbol{\sigma}}{\boldsymbol{d}}\right)^2 \quad (1)$$

Where σ is the standard deviation (nm) or peak width and *d* is the mean population diameter or peak in nm, thus the PDI is considered a dimensionless number determined by the square of the standard deviation ratio between the mean diameter of each population in the distribution (Nobbmann, 2015).

2.3.3. Ultraviolet and visible molecular absorption spectroscopy (UV-Vis)

The Ag-NPs were characterized by estimated size in an UV-Vis absorption spectrum in the spectrophotometer, UV-1601 PC Shimadzu model (Markham, CAN). 1 mL of the dispersion was inserted into a quartz cuvette with 10 mm optical path, then the cuvette was inserted into the spectrophotometer for reading and the wavelength range for scanning used was 290 nm to 700 nm.

2.4. Antimicrobial activity of silver nanoparticles

The antimicrobial activity of the Ag-NPs was performed according to the microdilution method determining the minimum inhibitory concentration (CLSI, 2012). In 96-well microplates solutions were added with concentrations of Ag-NPs varying from (0.060 to 61.75) $\mu g.mL^{-1}$, which were diluted in Mueller-Hinton agar inoculated with *Salmonella* serotypes (Table 1) in concentration of 10^5 $CFU.mL^{-1}$. After 48 h of incubation at 37 °C, it was checked which wells showed turbidity, observed in the Reader of the Elisa Expert Plus (BIOCHROM). MIC was the lowest concentration that inhibited bacterial multiplication. As

confirmation of inhibition of bacterial growth, aliquots of the positive and negative wells were plated in standard counting Agar (PCA) (HIMEDIA).

3. Results

3.1. Determination of the silver concentration in the dispersion

The concentration of Ag-NPs in the synthesis solution was 130 $\mu g \cdot mL^{-1}$. The ICP-OES is capable of simultaneously analyzing various elements and determines chemical elements in solution in the form of ions, with advantages in relation to other techniques lying in the limit of detection, rapidity of analysis and high isotopic capacity, since it is an analysis that determines the elementary form (Perkin Elmer, 2004; Wolf, 2005). Guzmam et al. (2012) also evaluated dispersions of Ag-NPs and found concentrations ranging from (112.72 to 595.44) $\mu g \cdot mL^{-1}$. Rogers et al. (2018) also assessed concentrations of Ag-NP dispersions using ICP-OES and found concentrations ranging from (0.54 to 960) $\mu g \cdot mL^{-1}$ of commercial products.

Some studies show that the concentration of silver in nanodispersions can be influenced by ionic strength, temperature used in the synthesis, silver oxidation state, concentration of the added reagents in the synthesis and size of the particles (Sotiriou et al., 2012; Zhang et al., 2011).

The concentration of silver found in this study is similar in comparison to the values found in the other studies, thus showing consistency with the methodology employed. The standardization of the method seems to be a crucial factor for the success of the production and the concentration of the dispersion.

3.2. Size and stability of silver nanoparticles (Dynamic Light Scattering (DLS) and Zeta Potential)

The size of the Ag-NPs determined by DLS and the zeta potential are shown in Table 2. The importance of characterizing size and stability of Ag-NPs is related to the ability of their ions to remain dispersed, considering the magnitude of negative or positive charges and the electrostatic repulsion between them, besides, these factors have a direct influence on the antimicrobial activity (Zhang et al., 2011; Sotiriou et al., 2012). The size of the Ag-NPs found of 4.7 ± 0.09 nm represented a volume of 99.4% of the population in the dispersion. A

population of secondary size was also found, representing a volume of 0.6% dispersion with size of 35.7 ± 2.12 nm. Losasso et al. (2014) found mean values of Ag-NPs ranging from (6 to 20) nm and found secondary values above 500 nm. Martínez-Castañón et al. (2008) found mean Ag-NP sizes varying from (7 to 89) nm. In this study, we found smaller nanoparticles sizes, which can represent a good standardization of the synthesis methodology and also present good antimicrobial action.

Table 2: Mean size (nm) and zeta potential (mV) of the synthesized Ag-NPs.

Size (nm)*	PDI	Zeta potential (mV)*
4.7 ± 0.09	0.004	-33.7 ± 11.8
35.7 ± 2.12	0.002	

* Average of three replicates.

Colloidal dispersions may present different populations that relate dispersion as mono or polydisperse characterized by the dispersion index (PDI). The dispersions may be classified as monodisperse, indicating homogeneity of the dispersion, as slightly polydisperse and very or highly polydisperse, indicating a certain heterogeneity of dispersion. PDI values below 0.1 classify dispersions as monodisperse, values between (0.1 to 0.4) classify dispersions as slightly polydisperse and values above 0.4 classify dispersions as very or highly polydisperse (Nobbmann and Morfesis, 2009; Bhattacharjee, 2016; Ardani et al., 2017). The PDI values obtained in this study classify the dispersion of Ag-NPs as monodisperse, considering that both populations showed PDI values <0.1. Nobbmann and Morfesis (2009) also evaluated metallic nanoparticles and found PDI values lower than 0.1 for main sizes of 76.9 nm, classifying their dispersion as monodisperse and without aggregations. Rogers et al. (2018) evaluated several silver-containing commercial sanitizing products and found average values ranging from (5 to 75) nm, all products being considered polydisperse dispersions and according to these same authors the dispersion index may vary if the stability of the nanostructures were changed.

The stability of Ag-NPs is measured by the Zeta potential. Agnihotri, Mukherji & Mukherji (2014) observed a Zeta potential value of -26.8 mV for Ag-NPs of size 5 nm, very close to that obtained in this study, and attributed it to high surface energies in the dispersed particles. The value of -33.7 mV found in this study indicates that the surfaces of the nanoparticles are negatively charged and show high dispersion stability. According to Sadowski et al. (2008), high stability is observed in dispersions with negative potential Zeta

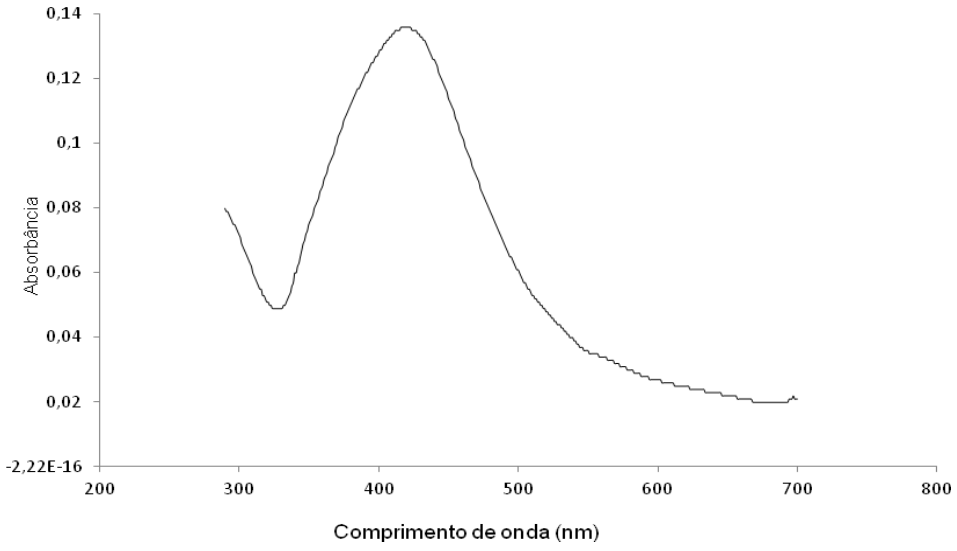
due to the electrostatic repulsion that occurs between the particles. Low Zeta potential negative values clearly indicate the instability of the dispersion, i.e., the closer to zero, the greater the instability (Sadowski et al., 2008).

However, over two weeks of storage an accumulation of Ag-NP aggregates, gray in color, was observed at the bottom, indicating loss of stability of the solution. The formation of Ag-Np aggregates occurs naturally over time (hours, weeks or even months) and is associated with a reduction in electrostatic repulsion which naturally attempts to decrease the total energy of the system, in addition to the concentration and type of stabilizing agent used. The presence of light also contributes to increased instability of the Ag-NPs, causing a change in color from yellow to gray and favoring the formation of aggregates. As an alternative to avoiding aggregations and loss of stability, current studies relate the synthesis of Ag-NPs with addition of stabilizers in order to increase the stability of these nanoparticles for commercial uses (Pinzaru et al.; 2018; Surmeneva et al.; 2017).

3.3. Particle size through molecular absorption spectroscopy ultraviolet and visible region (UV-Vis)

Ag-NPs were represented by a plasmonic surface at a peak near 400 nm (Figure 1).

Figure 1: Optical absorption spectrum in the UV-Vis region of chemically synthesized Ag-NPs .



Source: Authors.

UV-Vis spectroscopy is widely used to identify the presence of Ag-NPs, to determine

sizes and structural characteristics, such as spherical, cubic or triangular shapes. This technique is also related to the coloration of nanoparticles in dispersions in which colors between yellow and lighter shades of brown are absorbed in plasmic surface peaks around 400 nm (Rogers et al., 2018).

According to Solomon et al. (2007), the peak absorbance around 400 nm indicates that the nanoparticles have sizes ranging from (5 to 15) nm, which corroborates the DLS analysis performed in this study whereby most of the particles were about 4.7 nm. Moreover, the demonstrated absorbance peak also indicates that the Ag-NPs are spherical in shape. According to Sosa, Noguez and Barrera (2003), the study of the plasmonic surface associated with the chemical properties of the elements results in symmetrical characteristics and size in the nanoparticles.

Guzman, Dille and Godet (2012) synthesized and evaluated Ag-NPs using UV-Vis and found a similar plasmonic surface value of 418 nm and also considered dispersion as a spherical shape. Rogers et al. (2018) evaluated nine products containing Ag-NPs and found plasmonic surface values ranging from (398 to 420) nm in five of the products, while in 4 of them it was not possible to identify plasmonic surface due to the low concentration of Ag-NPs. Thus, the peak of absorbance depends on shape and size of the nanoparticle and can be displaced while surface interactions between the particles and between the suspension medium occur.

3.4. Antimicrobial activity of silver nanoparticles

The MIC of the Ag-NPs for the *Salmonella* serotypes evaluated are shown in Table 3. Four of the serotypes evaluated were found to have MICs of 11.8 $\mu g.mL^{-1}$ (Typhimurium, Schwarzengrund, Heidelberg and *Salmonella* infantis isolate 2), the Enteritidis serotype presented the lowest MIC of 4.7 $\mu g.mL^{-1}$ and *Salmonella* Infantis isolate 1 had the highest MIC of 27.7 $\mu g.mL^{-1}$ after 48 h of incubation.

Table 3: Minimum Inhibitory Concentration (MIC) of silver nanoparticles Ag-NPs on different serotypes of *Salmonella enterica.*

Bacteria / serotypes	**MIC (µg.mL^{-1})**
Salmonella Enteritidis	4.7
Salmonella Typhimurium	11.8
Salmonella Schwarzengrund	11.8
Salmonella Heidelberg	11.8
Salmonella Infantis 1	27.7
Salmonella Infantis 2	11.8

Source: Authors.

A difference in MICs between the *Salmonella* serotypes studied was observed. This fact indicates a different resistance profile among these, including between the two bacteria belonging to the Infantis serotype which showed contrast values. The MIC of 4.7 µg·mL^{-1} for *Salmonella* Enteritidis contrasts with the MIC of 16 µg·mL^{-1} for this same bacterium in a recent study by Omara et al. (2017), which was approximately 4 times higher, whereas for S. Typhimurium the observed MICs were similar (9 and 11.8) µg·mL^{-1}.

Although the *Salmonella* serotypes showed different resistance profiles, the MIC for the bacteria tested was considered lower than other studies with gram positive bacteria (Buszewski et al, 2016; Panácek et al, 2017), indicating that larger concentrations may have a good bactericidal effect. Few studies have been conducted regarding the action of Ag-NPs on the various *Salmonella* serotypes and the fact that their ability to transfer genes of resistance and pathogenicity is increasing means the use of Ag-NPs can be explored in further studies on this genus .

Losasso et al. (2014) compared the antibacterial effect of Ag-NPs on certain *Salmonella* serotypes and found that decimal reductions in bacterial populations may depend on the serotype and that 200 µg·mL^{-1} was the most effective dosing against the serotypes tested. Much lower values were observed by Zarei, Jamnejad and Khajehali (2014) who found a MIC of 3.12 µg·mL $^{-1}$ against *Salmonella* Typhimurium strains. Morones et al. (2005) evaluated the effect of different concentrations of silver nanoparticles on the growth of *Escherichia coli* and found MICs of 75 µg·mL^{-1}. Buszewski et al. (2016) found an MIC value of 6.25 µg·mL^{-1} for *E.coli*, which was lower than that found by previous authors. The latter authors also tested MICs for *Pseudomonas aeruginosa* and *Salmonella* Infantis, with MIC values of 25 and 6.25 µg·mL^{-1}, respectively. Some authors have stated that Ag-NPs are more

effective on gram negative bacteria than gram positive bacteria (Buszewski et al, 2016; Panácek et al, 2017). In contrast, recent studies have shown that the interaction of Ag-NPs with the bacterial surface depends on how their synthesis was performed and the type of coating used, considering that according to the bacterial structures these Ag-NPs can be coated, which will allow them to more easily permeate the plasma membrane of cells, regardless of whether they are gram positive or negative (Padmos et al., 2015; Zheng et al., 2018).

The shape, size, surface area and stability of Ag-NPs are deemed to be the main factors dictating bactericidal performance.Recent study (Omara et al., 2017) have shown results similar to those obtained in this study, in which reduced nanoparticle size may be related to the MICs. Flores et al. (2013) reported the effect of size on the MIC of Ag-NPs, where for 100 nm Ag-NPs the MIC was 104 $\mu g \cdot mL^{-1}$, and for those of size 5-20 nm, 12-25 $\mu g \cdot ML^{-1}$. One of the mechanisms behind the antibacterial activity of Ag-NPs, according to Baker et al. (2005), refers to its relation to the surface area. Particles of smaller size have a greater surface area ratio, which allows them to interact intimately with the microbial membranes. This interaction with the membrane compromises cellular respiration and leads to membrane rupture, causing loss of ATP, leading to cell death (Zhang and Chen, 2009).

Panacék et al. (2006), in a characterization study of Ag-NPs, synthesized nanoparticles with sizes between (25 and 450) nm. The authors observed that smaller nanoparticles had a greater bactericidal effect, confirming the fact that the antimicrobial effect of Ag-NPs is directly related to their size. In a similar study, Morones et al. (2005) characterized Ag-NPs and a satisfactory bactericidal effect was observed for nanoparticles of size 5 nm, which is in agreement with the result obtained in this study, in which the main size of the synthesized Ag-NPs was of 4.7 nm and being substantially small, has great potential as an antimicrobial agent.

The results obtained are consistent with those reported by other research papers. However, little is known about the toxicological information of Ag-NPs, which is of great importance for food safety. If these nanoparticles are to be used as a sanitizing agent in food industries in Brazil, permits must be granted by the existing legislative bodies. The main question to be posed is whether or not there is migration of Ag-NPs from the food processing surfaces to the final product. For this, further studies of risk analysis and proof of the absence of residual toxicological effects on humans are required.

4. Final Considerations

Characterization of Ag-NPs using UV-VIS and DLS confirmed the formation of the nanoparticles in colloidal dispersion. These nanoparticles had predominant average sizes of 4.7 nm, considered small and therefore efficient for antimicrobial activity. The synthesized Ag-NPs had a Zeta potential of -33.7 ± 11.8, which would indicate a dispersion of good thermodynamic stability. However, loss of stability was observed during storage, which can be solved with the addition of stabilizing substances during synthesis.

The Ag-NPs showed antimicrobial activity on all serotypes of *Salmonella* spp. evaluated, with different MIC values for the serotypes tested. Therefore, the use of Ag-NPs can be considered a strategy for bacterial control in food. It remains a challenge for researchers to produce nanoparticles which remain stable for commercial use. Moreover, further studies should be carried out to understand the mechanism of action of Ag-NPs on bacteria, including the genus *Salmonella*, considered one of the most significant in the food area.

As suggestions to for future work, it is possible to carry out the study with different stabilizing agents in the synthesis of Ag-NPs, also testing different concentrations of nanoparticles on *Salmonella* and also on other types of microorganisms that can cause problems for the food industry.

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